

Electric Supplementary Information (ESI)

Photo-triggered enzymatic degradation of biodegradable polymers

Yoshihiro Kikkawa,* Satoko Tanaka and Yasuo Norikane*

*National Institute of Advanced Industrial Science and Technology (AIST), Tsukuba Central 5, 1-1-1
Higashi, Tsukuba, Ibaraki 305-8565, Japan*

S1. Surface morphologies of thin films

S2. Enzymatic degradation of PLLA thin films by proteinase K

S3. FT-IR analyses of PLLA bilayer thin films

S4. Enzymatic degradation of PCL bilayer thin film by lipase

S5. Surface patterning by the combination of UV irradiation and enzymatic degradation

S1. Surface morphologies of thin films

To check the differences in the surface morphologies between pristine PLLA and bilayer thin film, AFM observation was performed for individual thin films. Figure S1A shows the AFM height image of PLLA amorphous thin film. In the case of the bilayer thin film, small crystallites with ca. 1 μm in width was observed on the whole surface, as shown in Figure S1B. After the irradiation of UV light (365 nm), the bilayer thin film was exposed to visible light (436 nm). AFM image in Figure S1C revealed that small crystallites were disappeared to show thin crystalline layers over 5 μm in size. This result suggests that the crystals of **Azo** on PLLA surface melts on the UV irradiation, re-crystallizes on the visible light irradiation and covers the PLLA surface.

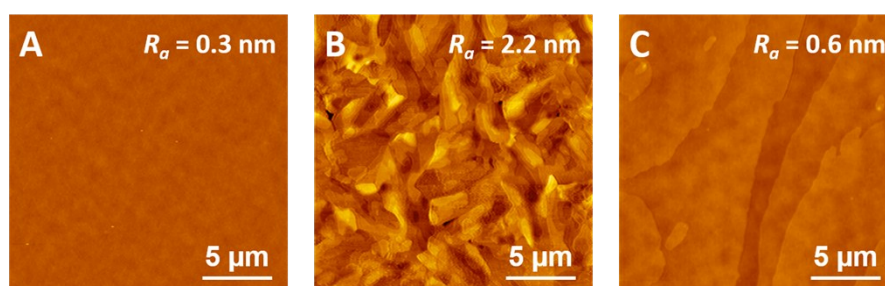


Figure S1 AFM height images of amorphous PLLA thin film (A), and bilayer thin film before UV(B) irradiation, and after successive irradiation of UV and visible light (C). The surface roughness (R_a) is indicated in the right corner of each image.

S2. Enzymatic degradation of PLLA thin films by proteinase K

Time-dependent erosion depth change due to enzymatic degradation by proteinase K was measured in the PLLA thin film (without **Azo**) pre-treated with UV-ozone by using AFM. Figure S2 shows a series of AFM images and cross-sectional profile of erosion depth (d), which was plotted as function of degradation time (red square plots in Figure 4). In each AFM image, left half side is eroded region, whereas the right side is non-eroded region. As the enzymatic degradation proceeded, the image contrast between left and right side became stronger, suggesting the erosion depth increases.

Erosion depth change during enzymatic degradation was also measured in pristine PLLA thin films with ca. 70 nm in thickness (without UV-ozone treatment). Figure S3 shows the plots of

time-dependent enzymatic erosion depth. The pristine PLLA thin film was eroded within 15 min (green lozenge plots).

Initially, it was suspected that molten **Azo** covering the PLLA surface prevents enzyme degradation. Therefore, after exposure to UV light (365 nm), the bilayer thin film was washed with aqueous ethanol (72% v/v) to remove molten **Azo**. Then, enzymatic degradation by proteinase K was performed. The enzymatic degradation rate (purple triangle plots) was almost identical to the result from Figure 4, in which the enzymatic degradation of PLLA was performed directly from the molten **Azo** surface. This result suggests that the molten **Azo** does not disturb the enzymatic degradation of PLLA. The enzymatic degradation rate of the bilayer thin film was almost half compared to the pristine PLLA thin film. In the bilayer thin film, UV ozone treatment was conducted for improving the adhesion of **Azo**. The result is consistent with our previous study,^{S1} namely, retardation of enzymatic degradation is introduced by the UV-ozone treatment of PLLA.

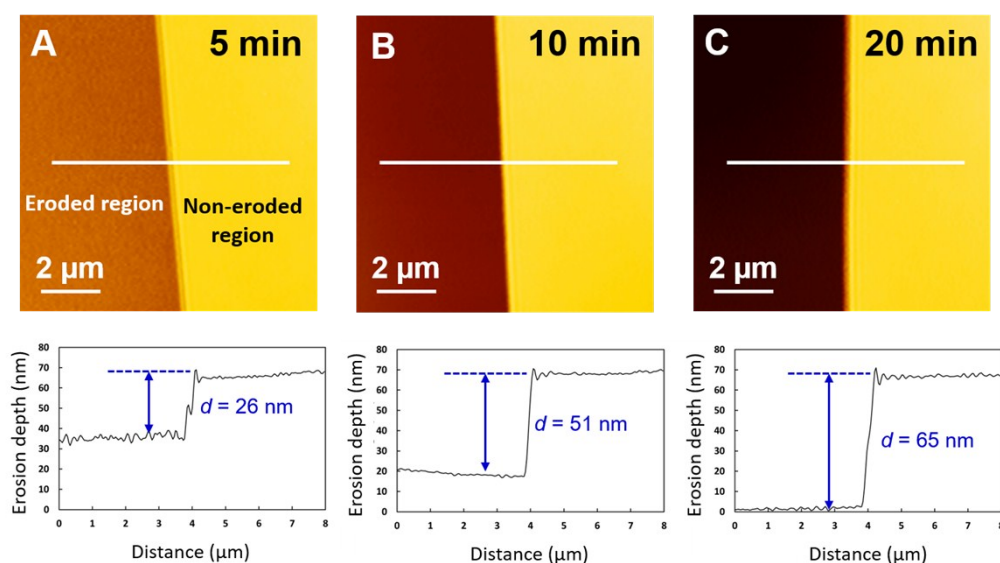


Figure S2 A series of AFM height images of PLLA thin film (pre-treated with UV-ozone) and cross-sectional profile of erosion depth (d) during the enzymatic degradation by proteinase K. It is evident that the d is increased with the process of enzymatic degradation.

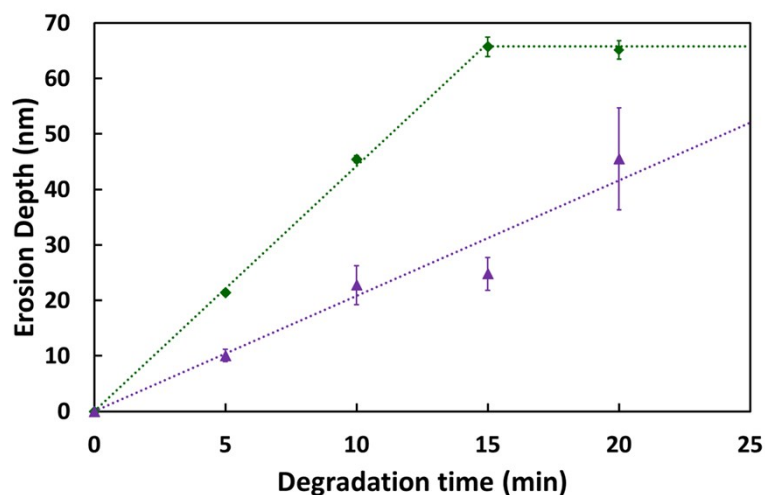


Figure S3 Time-dependent enzymatic erosion of pristine PLLA thin film (green lozenge plots) and bilayer thin film after the exposure to UV light and washing with ethanol aqueous solution (purple triangle plots).

S3. FT-IR analyses of PLLA bilayer thin films

FT-IR measurements (JASCO FT/IR420) of thin films were performed to confirm which component is covering the material surface. Either PLLA or **Azo** thin film was prepared on a KBr plate via spin cast method. The PLLA surface was treated with UV-ozone. In the FT-IR spectra, there are two prominent peaks when analyzing the bilayer thin film: the **Azo** (Figure S4A) showed the typical peaks at 2920 and 2851 cm^{-1} (C-H stretch) derived from decyloxy chains, whereas the PLLA (Figure S4B) showed the C=O stretching peak at 1753 cm^{-1} . These characteristic peaks can be used for the determination of which component is covering the surface of the thin film.

The bilayer thin film (Figure S4C) exhibited the spectrum, which was same as the superposition of two spectra in Figure S4A and S4B. After exposure to UV light (365 nm) for 1 min and following washing with EtOH aqueous solution (72%), the peaks derived from Azo was disappeared in Figure S4D, and the spectrum became identical to that of the PLLA in Figure S4B. Note that just washing of the bilayer thin film with EtOH aqueous solution (Figure S4E) did not change the characteristic peaks in the spectrum, namely the peak positions were same as Figure S4C. These results suggest that the PLLA surface is certainly covered with the **Azo**, which cannot be removed without UV irradiation. Therefore, the data in Figure 4 can be interpreted as follows: the **Azo** layer hinders the enzymatic degradation of PLLA layer because the proteinase K cannot reach

and attack the PLLA surface. Once the **Azo** is melted by the UV irradiation, the enzyme can approach to the PLLA surface to hydrolyze and erode the material.

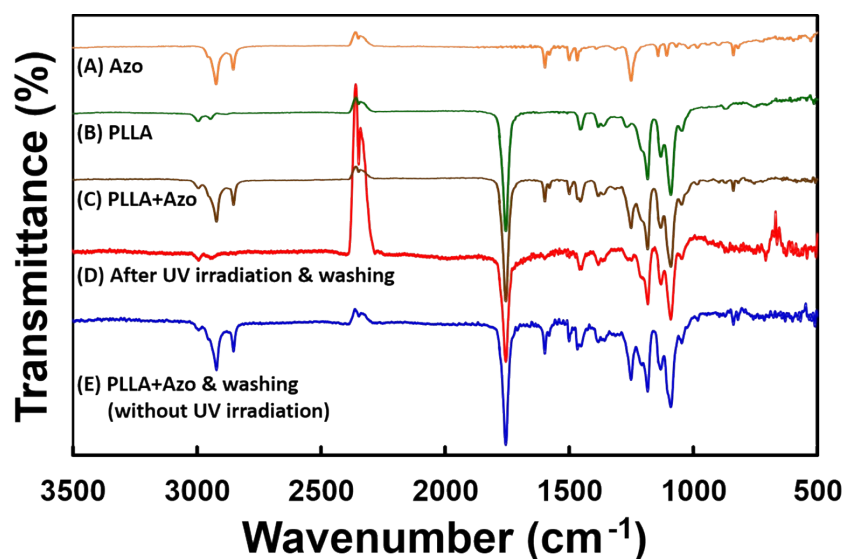


Figure S4 FT-IR spectra of (A) Azo, (B) PLLA pretreated with UV-ozone, (C) a bilayer thin film, (D) a bilayer thin film after the UV irradiation and washing with EtOH aq. solution, and (E) a bilayer thin film (without UV irradiation) washed with EtOH aq. solution. The spectra were normalized to the peak at either C-H or C=O stretch.

S4. Enzymatic degradation of PCL bilayer thin film by lipase

A PCL chloroform solution (1% w/v) was spin cast on a Si substrate, and the bilayer thin film was prepared as the same method in Scheme 1. Then, UV irradiation and following enzymatic degradation was performed by using from *Pseudomonas sp.* lipase (Sigma-Aldrich: 1 mg/mL in phosphate buffer pH = 7.4). The erosion depth was measured after the enzymatic degradation for 2h and 6h, as shown in Figure S5. The erosion rate of the bilayer thin film was almost identical to the UV-ozone treated PCL thin film (the data was introduced from our previous study^{S1}). This result suggests that the initiation of enzymatic degradation of PCL is controlled by the phase transition of **Azo** induced by the UV irradiation.

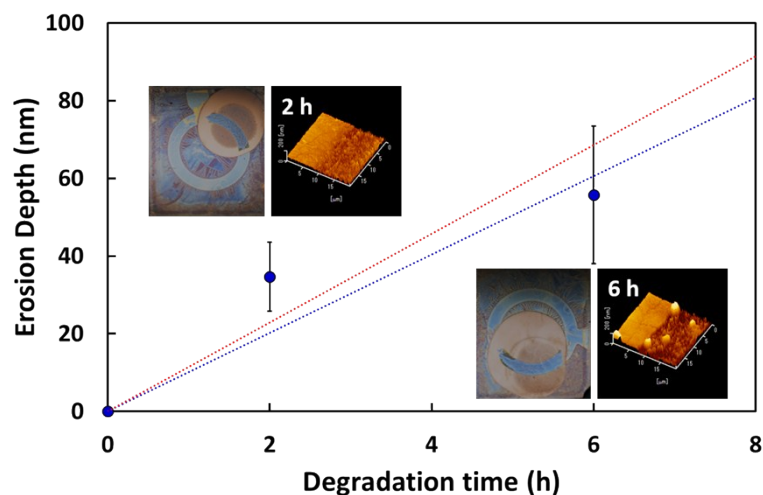


Figure S5 Time-course of enzymatic erosion of bilayer PCL thin film by lipase (blue circled plots). Inset: photographs of bilayer thin film after the UV exposure and enzymatic degradation, and AFM height images. The red line is corresponding to the degradation manner of UV-ozone treated PCL thin film in our previous study.^{S1}

S5. Surface patterning by the combination of UV irradiation and enzymatic degradation

UV irradiation under specific mask allowed to produce the patterned surface of **Azo** on PLLA (Figure S6A). Then, droplet of proteinase K solution was placed at the defined position, and enzymatic degradation was performed. Then, 5×5 circular holes (ca. 1.3 mm diameter) were created in the thin film (Figure S6B). Thus, combinations of phase structure control by UV irradiation and enzymatic degradation would provide the new environmentally friendly and mild lithographic technique.

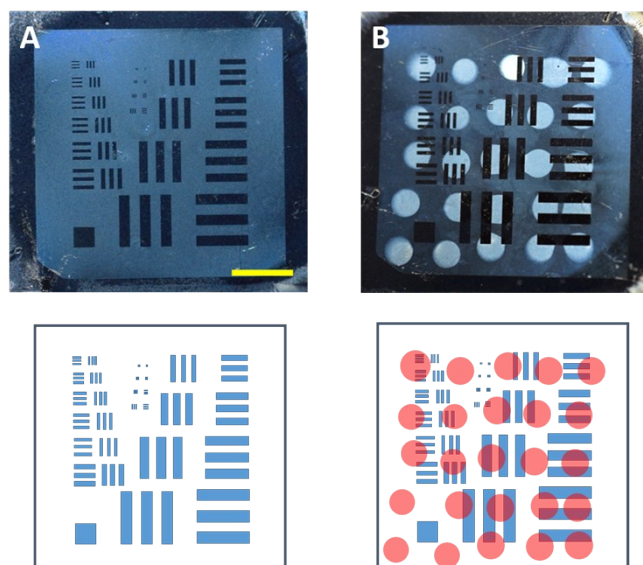


Figure S6 Demonstration of surface patterning of **Azo** by UV irradiation (365 nm) (A), and following enzymatic degradation of PLLA (B). Scale bar = 3 mm.

References

- S1 Y. Kikkawa, M. Fukuda, N. Ichikawa, A. Kashiwada, K. Matsuda, M. Kanesato and T. Hiraishi, *J. Mater. Chem. A*, 2013, **1**, 4667.